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PTO/SB/05 (12/97)
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jc534 U.S. PTO 01/27/98

UTILITY PATENT APPLICATION TRANSMITTAL

(Only for new nonprovisional applications under 37 CFR 1.53(b))

Attorney Docket No.	BOER 1059.1
Total Pages	
First Named Inventor or Application Identifier	Ulrich Martin et al.
Express Mail Label No.	EI828162331US

APPLICATION ELEMENTS

See MPEP chapter 600 concerning utility patent application contents.

ADDRESS TO: Assistant Commissioner for Patents, Box Patent Application, Washington, DC 20231

1. [X] Fee Transmittal Form *(Submit an original, and a duplicate for fee processing)*
2. [X] Specification [Total Pages 43] *(preferred arrangement set forth below)*
   - Descriptive title of the Invention
   - Cross References to Related Applications
   - Statement Regarding Fed sponsored R & D
   - Reference to Microfiche Appendix
   - Background of the Invention
   - Brief Summary of the Invention
   - Brief Description of the Drawings *(if filed)*
   - Detailed Description
   - Claim(s)
   - Abstract of the Disclosure
3. [X] Drawing(s) *(35 USC 113)* [Total Sheets 5]
4. Oath or Declaration [Total Pages 3]
   a. [X] Newly executed (original or copy)
   b. [ ] Copy from a prior application (37 CFR 1.63(d)) *(for continuation/divisional with Box 17 completed)* *[Note Box 5 below]*
      i. [ ] DELETION OF INVENTOR(S) Signed statement attached deleting inventor(s) named in the prior application, see 37 CFR 1.63(d)(2) and 1.33(b).
5. [X] Incorporation By Reference *(useable if Box 4b is checked)* The entire disclosure of the prior application, from which a copy of the oath or declaration is supplied under Box 4b, is considered as being part of the disclosure of the accompanying application and is hereby incorporated by reference therein.
6. [ ] Microfiche Computer Program *(Appendix)*
7. Nucleotide and/or Amino Acid Sequence Submission *(if applicable, all necessary)*
   a. [ ] Computer Readable Copy
   b. [X] Paper Copy (identical to computer copy)
   c. [ ] Statement verifying identity of above copies

ACCOMPANYING APPLICATION PARTS

8. [ ] Assignment Papers (cover sheet & document(s))
9. [ ] 37 CFR 3.73(b) Statement *(when there is an assignee)* [ ] Power of Attorney
10. [ ] English Translation Document *(if applicable)*
11. [ ] Information Disclosure Statement (IDS)/PTO-1449 [ ] Copies of IDS Citations
12. [X] Preliminary Amendment
13. [X] Return Receipt Postcard (MPEP 503) *(Should be specifically itemized)*
14. [ ] Small Entity Statement(s) [ ] Statement filed in prior application, Status still proper and desired
15. [ ] Certified Copy of Priority Document(s) *(if foreign priority is claimed)*
16. [X] Other: Copy of Published PCT Application

17. If a CONTINUING APPLICATION, *check appropriate box and supply the requisite information:*
[X] Continuation [ ] Divisional [ ] Continuation-in-part (CIP) of prior application No: PCT/US96/13152

18. CORRESPONDENCE ADDRESS

[ ] Customer Number or Bar Code Label [ ] (Insert Customer No. or Attach bar code label here) or [X] Correspondence address below

NAME	Norman D. Hanson				
	FELFE & LYNCH				
ADDRESS	805 Third Avenue				
CITY	New York	STATE	NY	ZIP CODE	10022
COUNTRY	USA	TELEPHONE	212-688-9200	FAX	212-838-3884

Burden Hour Statement: This form is estimated to take 0.2 hours to complete. Time will vary depending upon the needs of the individual case. Any comments on the amount of time you are required to complete this form should be sent to the Chief Information Officer, Patent and Trademark Office, Washington, DC 20231. DO NOT SEND FEES OR COMPLETED FORMS TO THIS ADDRESS. SEND TO: Assistant Commissioner for Patents, Box Patent Application, Washington, DC 20231.

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Docket No.: BOER 1059.1-PFF/NDH
Anticipated Art Group:
Date: January 27, 1998

Hon. Commissioner of Patents
and Trademarks
Washington, D.C. 20231

EI828162331US

S I R:

(X) This is a request for filing a Continuation application under 37 CFR 1.53(b)

of International application No. PCT/US96/13152 filed on 13 August 1996 which is a continuation-in-part of U.S. Application Serial No. 08/578,953 filed on December 27, 1997

Ulrich Martin et al.
(inventors)

for ANTI-SELECTIN ANTIBODIES FOR PREVENTION OF MULTIPLE ORGAN FAILURE AFTER POLYTRAUMA AND FOR PREVENTION OF ACUTE ORGAN DAMAGE AFTER EXTRACORPOREAL BLOOD CIRCULATION
(TITLE OF INVENTION)

ATTACHED IS A TRUE COPY OF SAID PRIOR APPLICATION AS FILED from the records of the Attorney of Record.

The filing fee is calculated below:
CLAIMS AS FILED, LESS ANY CLAIMS CANCELLED BY AMENDMENT BELOW

For	Number Filed	Number Extra	Rate		Basic fee ($790/395)
Total Claims.....	28	8	x $22/11	=	$176.00
Independent Claims.	3	0	x $82/41	=	$

( ) Multiple Dependent Claims - where applicable ($270/135)
( ) Foreign language text - where applicable ($130)
(X) Late Filing of Declaration ($130/$65)

TOTAL FILING FEE $ 1096.00

(X) The filing fee of $1096.00 is enclosed. In the event the enclosed check is unacceptable and/or insufficient to cover the required fees, please charge to account No. 06-0530.

Respectfully submitted,

FELFE & LYNCH

By _____
Norman D. Hanson
Reg. No. 30,946

805 Third Avenue
New York, New York 10022
(212) 688-9200
Enclosure

BOER 1059.1-PFF\NDH

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicants	:	Ulrich Martin et al.
Serial No.	:	Continuation of PCT/US96/13152
Filed	:	Concurrently herewith
For	:	ANTI-SELECTIN ANTIBODIES FOR PREVENTION OF MULTIPLE ORGAN FAILURE AFTER POLYTRAUMA AND FOR PREVENTION OF ACUTE ORGAN DAMAGE AFTER EXTRACORPOREAL BLOOD CIRCULATION

Hon. Commissioner of Patents
and Trademarks
Washington, D.C. 20231

PRELIMINARY AMENDMENT

S I R :

Prior to examination, please amend this application as follows:

IN THE SPECIFICATION

Prior to "FIELD OF THE INVENTION" add

-- This is a continuation of International Application PCT/US96/13152, with an international filing date of August 16, 1996, which is a continuation in part of U.S. application Serial No. 08/578,953, filed on December 27, 1995. Both applications are incorporated by reference in their entirety.

IN THE CLAIMS

Claim 7, line 1: change "1" to -- 2 --.

Claim 8, line 1: change "1" to -- 2 --.

Claim 27, line 1: change "22" to -- 24 --.

REMARKS

This application is filed as a continuation application which is permitted by 35 USC § 365(c) and 120. See MPEP 1895.

The specification has been amended to refer explicitly to the International Application from which priority is claimed. A copy of this published application is provided. Note that the US was designated.

Minor changes have been made to the claims. Entry of this amendment is requested.

Respectfully submitted,

FELFE & LYNCH

By ______________________
Norman D. Hanson
Reg. No. 30,946

805 Third Avenue
New York, New York 10022
(212) 688-9200

BOER 1059.1-PFF\NDH

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicants	:	Ulrich Martin et al.
Serial No.	:	Continuation of PCT/US96/13152
Filed	:	Concurrently herewith
For	:	ANTI-SELECTIN ANTIBODIES FOR PREVENTION OF MULTIPLE ORGAN FAILURE AFTER POLYTRAUMA AND FOR PREVENTION OF ACUTE ORGAN DAMAGE AFTER EXTRACORPOREAL BLOOD CIRCULATION

Hon. Commissioner of Patents
and Trademarks
Washington, D.C. 20231

LETTER RE SEQUENCES (37 CFR § 1.821(e)

Sir:

The sequences in this application are identical to the sequences filed in Serial No. 08/578,953 on January 20, 1998. A paper copy of that sequence listing is attached. Please insert this after page 26, cancelling pages 27-36. Note that SEQ ID NOS: 5 and 6 correspond to materials incorporated by reference in the subject application.

Please transfer the computer readable form of sequence information filed in Serial No. 08/578,953 on January 20, 1998, to this application.

The undersigned hereby declares that, to the best of his knowledge the paper copy submitted herewith, the paper copy submitted in 08/578,953 on January 20, 1998 and the computer

readable form of sequence information submitted in 08/578,953 on January 20, 1998 are all identical to each other. No new matter is presented.

Respectfully submitted,

FELFE & LYNCH

By _____
Norman D. Hanson
Reg. No. 30,946

805 Third Avenue
New York, New York 10022
(212) 688-9200

SEQUENCE LISTING

(1) GENERAL INFORMATION:

(i) APPLICANTS: MARTIN, Ulrich; HASELBECK, Anton; SCHUMACHER, Guenther; CO, Man S.

(ii) TITLE OF INVENTION: ANTI-L-SELECTIN ANTIBODIES FOR PREVENTION OF MULTIPLE ORGAN FAILURE AFTER POLYTRAUMA AND FOR PREVENTION OF ACUTE ORGAN DAMAGE AFTER EXTRACORPOREAL BLOOD CIRCULATION

(iii) NUMBER OF SEQUENCES: 6

(iv) CORRESPONDENCE ADDRESS:
- (A) ADDRESSEE: Felfe & Lynch
- (B) STREET: 805 Third Avenue
- (C) CITY: New York
- (D) STATE: New York
- (E) COUNTRY: U.S.A.
- (F) ZIP: 10022

(v) COMPUTER READABLE FORM:
- (A) MEDIUM TYPE: 3.5" Computer Disk
- (B) COMPUTER: IBM PC compatible
- (C) OPERATING SYSTEM: PC-DOS/MS-DOS
- (D) SOFTWARE: ASCII, WordPerfect 5.1

(vi) CURRENT APPLICATION DATA:
- (A) APPLICATION NUMBER: 08/578,953
- (B) FILING DATE: 27-Dec-95
- (C) CLASSIFICATION: 530

(vii) PRIOR APPLICATION DATA:
- (A) APPLICATION NUMBER: EP 95 112 895.8
- (B) FILING DATE: 17-Aug-95
- (A) APPLICATION NUMBER: EP 95 114 969.9
- (B) FILING DATE: 19-Sep-95

(viii) ATTORNEY/AGENT INFORMATION
- (A) NAME: Hanson, Norman D.
- (B) REGISTRATION NUMBER: 30,946
- (C) REFERENCE/DOCKET NUMBER: BOER 1059-PFF/NDH/SLH

(ix) TELECOMMUNICATION INFORMATION
- (A) TELEPHONE: (212) 688-9200
- (B) TELEFAX: (212) 838-3884

(2) INFORMATION FOR SEQ ID NO: 1:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 654 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: double
(D) TOPOLOGY: linear

(ii) MOLECULE TYPE: cDNA

(ix) FEATURE:
(A) NAME/KEY: CDS
(B) LOCATION: 1,,654

(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 1:

```
GAC ATT CAG ATG ACC CAA TCT CCG AGC TCT TTG TCT GCG TCT GTA GGG  48
Asp Ile Gln Met Thr Gln Ser Pro Ser Ser Leu Ser Ala Ser Val Gly
 1               5                  10                  15

GAT AGG GTC ACT ATC ACC TGC AAG GCC AGC CAA AGT GTT GAT TAT GAT  96
Asp Arg Val Thr Ile Thr Cys Lys Ala Ser Gln Ser Val Asp Tyr Asp
            20                  25                  30

GGT GAT AGT TAT ATG AAC TGG TAC CAA CAG AAA CCA GGA AAG GCA CCC  144
Gly Asp Ser Tyr Met Asn Trp Tyr Gln Gln Lys Pro Gly Lys Ala Pro
        35                  40                  45

AAG CTT CTC ATC TAT GCT GCA TCC AAC CTA GAA TCT GGT ATC CCA TCC  192
Lys Leu Leu Ile Tyr Ala Ala Ser Asn Leu Glu Ser Gly Ile Pro Ser
    50                  55                  60

AGG TTT AGT GGC AGT GGG TCT GGG ACA GAC TTC ACC CTC ACC ATC TCT  240
Arg Phe Ser Gly Ser Gly Ser Gly Thr Asp Phe Thr Leu Thr Ile Ser
65                  70                  75                  80

TCT CTG CAG CCG GAG GAT TTC GCA ACC TAT TAC TGT CAG CAA AGT AAT  288
Ser Leu Gln Pro Glu Asp Phe Ala Thr Tyr Tyr Cys Gln Gln Ser Asn
                85                  90                  95

GAA GAT CCG TGG ACG TTC GGT CAA GGC ACC AAG GTG GAA ATC AAA CGA  336
Glu Asp Pro Trp Thr Phe Gly Gln Gly Thr Lys Val Glu Ile Lys Arg
            100                 105                 110

ACT GTG GCT GCA CCA TCT GTC TTC ATC TTC CCG CCA TCT GAT GAG CAG  384
Thr Val Ala Ala Pro Ser Val Phe Ile Phe Pro Pro Ser Asp Glu Gln
        115                 120                 125

TTG AAA TCT GGA ACT GCC TCT GTT GTG TGC CTG CTG AAT AAC TTC TAT  432
Leu Lys Ser Gly Thr Ala Ser Val Val Cys Leu Leu Asn Asn Phe Tyr
    130                 135                 140
```

```
CCC AGA GAG GCC AAA GTA CAG TGG AAG GTG GAT AAC GCC CTC CAA TCG  480
Pro Arg Glu Ala Lys Val Gln Trp Lys Val Asp Asn Ala Leu Gln Ser
145                 150                 155                 160

GGT AAC TCC CAG GAG AGT GTC ACA GAG CAG GAC AGC AAG GAC AGC ACC  528
Gly Asn Ser Gln Glu Ser Val Thr Glu Gln Asp Ser Lys Asp Ser Thr
                165                 170                 175

TAC AGC CTC AGC AGC ACC CTG ACG CTG AGC AAA GCA GAC TAC GAG AAA  576
Tyr Ser Leu Ser Ser Thr Leu Thr Leu Ser Lys Ala Asp Tyr Glu Lys
            180                 185                 190

CAC AAA GTC TAC GCC TGC GAA GTC ACC CAT CAG GGC CTG AGC TCG CCC  624
His Lys Val Tyr Ala Cys Glu Val Thr His Gln Gly Leu Ser Ser Pro
        195                 200                 205

GTC ACA AAG AGC TTC AAC AGG GGA GAG TGT                          654
Val Thr Lys Ser Phe Asn Arg Gly Glu Cys
    210                 215
```

(2) INFORMATION FOR SEQ ID NO: 2:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 218
(B) TYPE: amino acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear

(ii) MOLECULE TYPE: protein

(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 2:

```
Asp Ile Gln Met Thr Gln Ser Pro Ser Ser Leu Ser Ala Ser Val Gly
 1               5                  10                  15

Asp Arg Val Thr Ile Thr Cys Lys Ala Ser Gln Ser Val Asp Tyr Asp
            20                  25                  30

Gly Asp Ser Tyr Met Asn Trp Tyr Gln Gln Lys Pro Gly Lys Ala Pro
        35                  40                  45

Lys Leu Leu Ile Tyr Ala Ala Ser Asn Leu Glu Ser Gly Ile Pro Ser
    50                  55                  60

Arg Phe Ser Gly Ser Gly Ser Gly Thr Asp Phe Thr Leu Thr Ile Ser
65                  70                  75                  80

Ser Leu Gln Pro Glu Asp Phe Ala Thr Tyr Tyr Cys Gln Gln Ser Asn
                85                  90                  95

Glu Asp Pro Trp Thr Phe Gly Gln Gly Thr Lys Val Glu Ile Lys Arg
            100                 105                 110
```

```
Thr Val Ala Ala Pro Ser Val Phe Ile Phe Pro Pro Ser Asp Glu Gln
        115                 120                 125

Leu Lys Ser Gly Thr Ala Ser Val Val Cys Leu Leu Asn Asn Phe Tyr
    130                 135                 140

Pro Arg Glu Ala Lys Val Gln Trp Lys Val Asp Asn Ala Leu Gln Ser
145                 150                 155                 160

Gly Asn Ser Gln Glu Ser Val Thr Glu Gln Asp Ser Lys Asp Ser Thr
                165                 170                 175

Tyr Ser Leu Ser Ser Thr Leu Thr Leu Ser Lys Ala Asp Tyr Glu Lys
            180                 185                 190

His Lys Val Tyr Ala Cys Glu Val Thr His Gln Gly Leu Ser Ser Pro
        195                 200                 205

Val Thr Lys Ser Phe Asn Arg Gly Glu Cys
    210                 215
```

(2) INFORMATION FOR SEQ ID NO: 3:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 1329 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: double
(D) TOPOLOGY: linear

(ii) MOLECULE TYPE: cDNA

(ix) FEATURE:
(A) NAME/KEY: CDS
(B) LOCATION:1,,1329

(ix) FEATURE:
(A) NAME/KEY: mat_peptide
(B) LOCATION:1

(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 3:

```
GAA GTG CAA CTG GTG GAG TCT GGG GGA GGC TTA GTG CAG CCT GGA GGA  48
Glu Val Gln Leu Val Glu Ser Gly Gly Gly Leu Val Gln Pro Gly Gly
  1               5                  10                  15

AGC TTG AGA CTC TCC TGT GCA GCC TCT GGA TTC ACT TTC AGT ACC TAT  96
Ser Leu Arg Leu Ser Cys Ala Ala Ser Gly Phe Thr Phe Ser Thr Tyr
            20                  25                  30

GCC ATG TCT TGG GTT CGC CAG GCT CCA GGG AAG GGA CTC GAG TGG GTC  144
Ala Met Ser Trp Val Arg Gln Ala Pro Gly Lys Gly Leu Glu Trp Val
        35                  40                  45
```

```
GCA TCC ATT AGT ACT GGT GGT AGC ACC TAC TAT CCA GAC AGT GTG AAG  192
Ala Ser Ile Ser Thr Gly Gly Ser Thr Tyr Tyr Pro Asp Ser Val Lys
     50                  55                  60

GGC CGA TTC ACC ATC TCC AGA GAT AAT GCC AAG AAC ACC CTG TAC CTG  240
Gly Arg Phe Thr Ile Ser Arg Asp Asn Ala Lys Asn Thr Leu Tyr Leu
 65                  70                  75                  80

CAA ATG AAT TCT CTG AGG GCT GAG GAC ACG GCC GTG TAT TAC TGT GCA  288
Gln Met Asn Ser Leu Arg Ala Glu Asp Thr Ala Val Tyr Tyr Cys Ala
                 85                  90                  95

AGA GAC TAT GAC GGG TAT TTT GAC TAC TGG GGC CAA GGC ACC CTG GTC  336
Arg Asp Tyr Asp Gly Tyr Phe Asp Tyr Trp Gly Gln Gly Thr Leu Val
            100                 105                 110

ACA GTC TCC TCA GCT TCC ACC AAG GGC CCA TCC GTC TTC CCC CTG GCG  384
Thr Val Ser Ser Ala Ser Thr Lys Gly Pro Ser Val Phe Pro Leu Ala
        115                 120                 125

CCC TGC TCC AGG AGC ACC TCC GAG AGC ACA GCC GCC CTG GGC TGC CTG  432
Pro Cys Ser Arg Ser Thr Ser Glu Ser Thr Ala Ala Leu Gly Cys Leu
    130                 135                 140

GTC AAG GAC TAC TTC CCC GAA CCG GTG ACG GTG TCG TGG AAC TCA GGC  480
Val Lys Asp Tyr Phe Pro Glu Pro Val Thr Val Ser Trp Asn Ser Gly
145                 150                 155                 160

GCC CTG ACC AGC GGC GTG CAC ACC TTC CCG GCT GTC CTA CAG TCC TCA  528
Ala Leu Thr Ser Gly Val His Thr Phe Pro Ala Val Leu Gln Ser Ser
                165                 170                 175

GGA CTC TAC TCC CTC AGC AGC GTG GTG ACC GTG CCC TCC AGC AGC TTG  576
Gly Leu Tyr Ser Leu Ser Ser Val Val Thr Val Pro Ser Ser Ser Leu
            180                 185                 190

GGC ACG AAG ACC TAC ACC TGC AAC GTA GAT CAC AAG CCC AGC AAC ACC  624
Gly Thr Lys Thr Tyr Thr Cys Asn Val Asp His Lys Pro Ser Asn Thr
        195                 200                 205

AAG GTG GAC AAG AGA GTT GAG TCC AAA TAT GGT CCC CCA TGC CCA TCA  672
Lys Val Asp Lys Arg Val Glu Ser Lys Tyr Gly Pro Pro Cys Pro Ser
    210                 215                 220

TGC CCA GCA CCT GAG TTC CTG GGG GGA CCA TCA GTC TTC CTG TTC CCC  720
Cys Pro Ala Pro Glu Phe Leu Gly Gly Pro Ser Val Phe Leu Phe Pro
225                 230                 235                 240

CCA AAA CCC AAG GAC ACT CTC ATG ATC TCC CGG ACC CCT GAG GTC ACG  768
Pro Lys Pro Lys Asp Thr Leu Met Ile Ser Arg Thr Pro Glu Val Thr
                245                 250                 255
```

```
TGC GTG GTG GTG GAC GTG AGC CAG GAA GAC CCC GAG GTC CAG TTC AAC  816
Cys Val Val Val Asp Val Ser Gln Glu Asp Pro Glu Val Gln Phe Asn
            260                 265                 270

TGG TAC GTG GAT GGC GTG GAG GTG CAT AAT GCC AAG ACA AAG CCG CGG  864
Trp Tyr Val Asp Gly Val Glu Val His Asn Ala Lys Thr Lys Pro Arg
        275                 280                 285

GAG GAG CAG TTC AAC AGC ACG TAC CGT GTG GTC AGC GTC CTC ACC GTC  912
Glu Glu Gln Phe Asn Ser Thr Tyr Arg Val Val Ser Val Leu Thr Val
    290                 295                 300

CTG CAC CAG GAC TGG CTG AAC GGC AAG GAG TAC AAG TGC AAG GTC TCC  960
Leu His Gln Asp Trp Leu Asn Gly Lys Glu Tyr Lys Cys Lys Val Ser
305                 310                 315                 320

AAC AAA GGC CTC CCG TCC TCC ATC GAG AAA ACC ATC TCC AAA GCC AAA 1008
Asn Lys Gly Leu Pro Ser Ser Ile Glu Lys Thr Ile Ser Lys Ala Lys
                325                 330                 335

GGG CAG CCC CGA GAG CCA CAG GTG TAC ACC CTG CCC CCA TCC CAG GAG 1056
Gly Gln Pro Arg Glu Pro Gln Val Tyr Thr Leu Pro Pro Ser Gln Glu
            340                 345                 350

GAG ATG ACC AAG AAC CAG GTC AGC CTG ACC TGC CTG GTC AAA GGC TTC 1104
Glu Met Thr Lys Asn Gln Val Ser Leu Thr Cys Leu Val Lys Gly Phe
        355                 360                 365

TAC CCC AGC GAC ATC GCC GTG GAG TGG GAG AGC AAT GGG CAG CCG GAG 1152
Tyr Pro Ser Asp Ile Ala Val Glu Trp Glu Ser Asn Gly Gln Pro Glu
    370                 375                 380

AAC AAC TAC AAG ACC ACG CCT CCC GTG CTG GAC TCC GAC GGC TCC TTC 1200
Asn Asn Tyr Lys Thr Thr Pro Pro Val Leu Asp Ser Asp Gly Ser Phe
385                 390                 395                 400

TTC CTC TAC AGC AGG CTA ACC GTG GAC AAG AGC AGG TGG CAG GAG GGG 1248
Phe Leu Tyr Ser Arg Leu Thr Val Asp Lys Ser Arg Trp Gln Glu Gly
                405                 410                 415

AAT GTC TTC TCA TGC TCC GTG ATG CAT GAG GCT CTG CAC AAC CAC TAC 1296
Asn Val Phe Ser Cys Ser Val Met His Glu Ala Leu His Asn His Tyr
            420                 425                 430

ACA CAG AAG AGC CTC TCC CTG TCT CTG GGT AAA                     1329
Thr Gln Lys Ser Leu Ser Leu Ser Leu Gly Lys
        435                 440
```

(2) INFORMATION FOR SEQ ID NO: 4:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 443
(B) TYPE: amino acid
(C) STRANDEDNESS: double
(D) TOPOLOGY: linear

(ii) MOLECULE TYPE: protein

(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 4:

```
Glu Val Gln Leu Val Glu Ser Gly Gly Gly Leu Val Gln Pro Gly Gly
 1               5                  10                  15

Ser Leu Arg Leu Ser Cys Ala Ala Ser Gly Phe Thr Phe Ser Thr Tyr
            20                  25                  30

Ala Met Ser Trp Val Arg Gln Ala Pro Gly Lys Gly Leu Glu Trp Val
        35                  40                  45

Ala Ser Ile Ser Thr Gly Gly Ser Thr Tyr Tyr Pro Asp Ser Val Lys
    50                  55                  60

Gly Arg Phe Thr Ile Ser Arg Asp Asn Ala Lys Asn Thr Leu Tyr Leu
65                  70                  75                  80

Gln Met Asn Ser Leu Arg Ala Glu Asp Thr Ala Val Tyr Tyr Cys Ala
                85                  90                  95

Arg Asp Tyr Asp Gly Tyr Phe Asp Tyr Trp Gly Gln Gly Thr Leu Val
            100                 105                 110

Thr Val Ser Ser Ala Ser Thr Lys Gly Pro Ser Val Phe Pro Leu Ala
        115                 120                 125

Pro Cys Ser Arg Ser Thr Ser Glu Ser Thr Ala Ala Leu Gly Cys Leu
    130                 135                 140

Val Lys Asp Tyr Phe Pro Glu Pro Val Thr Val Ser Trp Asn Ser Gly
145                 150                 155                 160

Ala Leu Thr Ser Gly Val His Thr Phe Pro Ala Val Leu Gln Ser Ser
                165                 170                 175

Gly Leu Tyr Ser Leu Ser Ser Val Val Thr Val Pro Ser Ser Ser Leu
            180                 185                 190

Gly Thr Lys Thr Tyr Thr Cys Asn Val Asp His Lys Pro Ser Asn Thr
        195                 200                 205

Lys Val Asp Lys Arg Val Glu Ser Lys Tyr Gly Pro Pro Cys Pro Ser
    210                 215                 220
```

```
Cys Pro Ala Pro Glu Phe Leu Gly Gly Pro Ser Val Phe Leu Phe Pro
225                 230                 235                 240

Pro Lys Pro Lys Asp Thr Leu Met Ile Ser Arg Thr Pro Glu Val Thr
                245                 250                 255

Cys Val Val Val Asp Val Ser Gln Glu Asp Pro Glu Val Gln Phe Asn
            260                 265                 270

Trp Tyr Val Asp Gly Val Glu Val His Asn Ala Lys Thr Lys Pro Arg
        275                 280                 285

Glu Glu Gln Phe Asn Ser Thr Tyr Arg Val Val Ser Val Leu Thr Val
    290                 295                 300

Leu His Gln Asp Trp Leu Asn Gly Lys Glu Tyr Lys Cys Lys Val Ser
305                 310                 315                 320

Asn Lys Gly Leu Pro Ser Ser Ile Glu Lys Thr Ile Ser Lys Ala Lys
                325                 330                 335

Gly Gln Pro Arg Glu Pro Gln Val Tyr Thr Leu Pro Pro Ser Gln Glu
            340                 345                 350

Glu Met Thr Lys Asn Gln Val Ser Leu Thr Cys Leu Val Lys Gly Phe
        355                 360                 365

Tyr Pro Ser Asp Ile Ala Val Glu Trp Glu Ser Asn Gly Gln Pro Glu
    370                 375                 380

Asn Asn Tyr Lys Thr Thr Pro Pro Val Leu Asp Ser Asp Gly Ser Phe
385                 390                 395                 400

Phe Leu Tyr Ser Arg Leu Thr Val Asp Lys Ser Arg Trp Gln Glu Gly
                405                 410                 415

Asn Val Phe Ser Cys Ser Val Met His Glu Ala Leu His Asn His Tyr
            420                 425                 430

Thr Gln Lys Ser Leu Ser Leu Ser Leu Gly Lys
        435                 440
```

(2) INFORMATION FOR SEQ ID NO: 5:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 220 amino acid residues
(B) TYPE: amino acid
(D) TOPOLOGY: linear

(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 5:

```
Asp Ile Gln Met Thr Gln Ser Pro Ser Thr Leu Ser Ala Ser Val Gly
1               5                  10                  15

Asp Arg Val Thr Ile Thr Cys Lys Ser Ser Gln Ser Leu Leu Asn Ser
            20                  25                  30

Ser Asn Gln Lys Asn Tyr Leu Ala Trp Tyr Gln Gln Lys Pro Gly Lys
        35                  40                  45

Ala Pro Lys Leu Leu Val Tyr Phe Ala Ser Thr Arg Glu Ser Gly Val
    50                  55                  60

Pro Asp Arg Phe Ile Gly Ser Gly Ser Gly Thr Asp Phe Thr Leu Thr
65                  70                  75                  80

Ile Ser Ser Leu Gln Pro Glu Asp Phe Ala Thr Tyr Phe Cys His Gln
                85                  90                  95

His Tyr Ser Thr Pro Leu Thr Phe Gly Gln Gly Thr Lys Val Glu Val
            100                 105                 110

Lys Arg Thr Val Ala Ala Pro Ser Val Phe Ile Phe Pro Pro Ser Asp
        115                 120                 125

Glu Gln Leu Lys Ser Gly Thr Ala Ser Val Val Cys Leu Leu Asn Asn
    130                 135                 140

Phe Tyr Pro Arg Glu Ala Lys Val Gln Trp Lys Val Asp Asn Ala Leu
145                 150                 155                 160

Gln Ser Gly Asn Ser Gln Glu Ser Val Thr Glu Gln Asp Ser Lys Asp
                165                 170                 175

Ser Thr Tyr Ser Leu Ser Ser Thr Leu Thr Leu Ser Lys Ala Asp Tyr
            180                 185                 190

Glu Lys His Lys Val Tyr Ala Cys Glu Val Thr His Gln Gly Leu Ser
        195                 200                 205

Ser Pro Val Thr Lys Ser Phe Asn Arg Gly Glu Cys
    210                 215                 220
```

(2) INFORMATION FOR SEQ ID NO: 6:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 448 amino acid residues
(B) TYPE: amino acid
(D) TOPOLOGY: linear

(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 6:

```
Gln Val Gln Leu Val Gln Ser Gly Ala Glu Val Lys Lys Pro Gly Ser
1               5                   10                  15

Ser Val Lys Val Ser Cys Lys Ala Ser Gly Tyr Thr Phe Thr Ser Tyr
            20                  25                  30

Val Met His Trp Val Arg Gln Ala Pro Gly Gln Gly Leu Glu Trp Ile
        35                  40                  45

Gly Tyr Ile Tyr Pro Tyr Asn Asp Gly Thr Lys Tyr Asn Glu Lys Phe
    50                  55                  60

Lys Gly Arg Val Thr Ile Thr Ser Asp Glu Ser Thr Asn Thr Ala Tyr
65                  70                  75                  80

Met Glu Leu Ser Ser Leu Arg Ser Glu Asp Thr Ala Val Tyr Tyr Cys
                85                  90                  95

Ala Arg Glu Glu Tyr Gly Asn Tyr Val Arg Tyr Phe Asp Val Trp Gly
            100                 105                 110

Gln Gly Thr Leu Val Thr Val Ser Ser Ala Ser Thr Lys Gly Pro Ser
        115                 120                 125

Val Phe Pro Leu Ala Pro Cys Ser Arg Ser Thr Ser Glu Ser Thr Ala
    130                 135                 140

Ala Leu Gly Cys Leu Val Lys Asp Tyr Phe Pro Glu Pro Val Thr Val
145                 150                 155                 160

Ser Trp Asn Ser Gly Ala Leu Thr Ser Gly Val His Thr Phe Pro Ala
                165                 170                 175

Val Leu Gln Ser Ser Gly Leu Tyr Ser Leu Ser Ser Val Val Thr Val
            180                 185                 190

Pro Ser Ser Ser Leu Gly Thr Lys Thr Tyr Thr Cys Asn Val Asp His
        195                 200                 205

Lys Pro Ser Asn Thr Lys Val Asp Lys Arg Val Glu Ser Lys Tyr Gly
    210                 215                 220

Pro Pro Cys Pro Ser Cys Pro Ala Pro Glu Phe Leu Gly Gly Pro Ser
225                 230                 235                 240

Val Phe Leu Phe Pro Pro Lys Pro Lys Asp Thr Leu Met Ile Ser Arg
                245                 250                 255

Thr Pro Glu Val Thr Cys Val Val Val Asp Val Ser Gln Glu Asp Pro
            260                 265                 270

Glu Val Gln Phe Asn Trp Tyr Val Asp Gly Val Glu Val His Asn Ala
        275                 280                 285
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Lys Thr Lys Pro Arg Glu Glu Gln Phe Asn Ser Thr Tyr Arg Val Val
290 295 300

Ser Val Leu Thr Val Leu His Gln Asp Trp Leu Asn Gly Lys Glu Tyr
305 310 315 320

Lys Cys Lys Val Ser Asn Lys Gly Leu Pro Ser Ser Ile Glu Lys Thr
325 330 335

Ile Ser Lys Ala Lys Gly Gln Pro Arg Glu Pro Gln Val Tyr Thr Leu
340 345 350

Pro Pro Ser Gln Glu Glu Met Thr Lys Asn Gln Val Ser Leu Thr Cys
355 360 365

Leu Val Lys Gly Phe Tyr Pro Ser Asp Ile Ala Val Glu Trp Glu Ser
370 375 380

Asn Gly Gln Pro Glu Asn Asn Tyr Lys Thr Thr Pro Pro Val Leu Asp
385 390 395 400

Ser Asp Gly Ser Phe Phe Leu Tyr Ser Arg Leu Thr Val Asp Lys Ser
405 410 415

Arg Trp Gln Glu Gly Asn Val Phe Ser Cys Ser Val Met His Glu Ala
420 425 430

Leu His Asn His Tyr Thr Gln Lys Ser Leu Ser Leu Ser Leu Gly Lys
435 440 445

TITLE

ANTI-SELECTIN ANTIBODIES FOR PREVENTION OF MULTIPLE ORGAN FAILURE AFTER POLYTRAUMA AND FOR PREVENTION OF ACUTE ORGAN DAMAGE AFTER EXTRACORPOREAL BLOOD CIRCULATION

FIELD OF THE INVENTION

This invention relates to the use of anti-selectin antibodies for the prevention of multiple organ failure associated with polytrauma and for the prevention of acute organ damage associated with extracorporeal blood circulation. Especially preferred are antibodies to E-selectin, L-selectin, and/or P-selectin.

BACKGROUND OF THE INVENTION

A polytrauma is understood as an injury of a number of tissues (bones or soft tissues). In a polytraumatic event, mediator systems (e.g. cytokines, arachidonic acid products, oxygen radicals, proteases) as well as leukocytes and other cells are activated. This can lead to secondary organ damage (e.g. destruction of tissue structures by liberated proteases). This secondary organ damage can occur in the whole body independently of the site of the primary trauma.

A polytrauma may also be associated with hemorrhagic shock. Hemorrhagic shock is understood as a shock which is characterized by a rapid and substantial loss of blood towards the inside or outside. At present, hemorrhagic shock can be treated successfully by intensive medical therapy, especially by volume substitution and blood transfusion. The combination of hemorrhagic shock and trauma is denoted hemorrhagic-traumatic shock. In contrast to pure hemorrhagic shock, there is, at present, no specific therapy for traumatic or

hemorrhagic-traumatic shock and no prophylaxis at all for later organ failure after polytrauma.

Multiple organ failure (MOF) is a severe problem which often occurs after polytraumas. The more organs affected, the higher the mortality. Organs which can fail are the heart, lung, kidney, liver, stomach, intestinal system and central nervous system. Although in recent years it has been possible to reduce the very high mortality of trauma patients to about 20 % by improvements in rescue service and emergency medicine, so far there is no specific therapy for organ failure.

Marzi et al., J. Trauma 35: 110-119 (1993) discloses that superoxide dismutase can be given 24 hours after trauma. However, the results are not unequivocal and merely show a trend towards a partial improvement. A substantial reduction in mortality and MOF was, however, not observed. Mileski, W.J. et al., Surgery 108: 206-212 (1990) discloses that the binding of neutrophils or their aggregation contributes substantially to the development of hemorrhagic shock after organ damage. In this case, experimental animals were given anti-CD18 antibodies immediately after a 90 minute phase of hemorrhagic shock. Therapeutic methods for treatment of multiple organ failure after polytrauma is not described by Mileski. Vedder N.B. et al., Surgery 106 (1989) 509-516 also propose the use of anti-CD18 antibodies to treat hemorrhagic shock.

Selectins, such as L, E, and P-selectin have been found to be associated with tissue damage during the course of ischemia and reperfusion. Neutrophils play an important role in this connection. It is assumed that selectin is required for the recruitment of neutrophils. Apparently L-selectin is necessary for the complete development of damage in skeletal muscle as well as in the lung (Seekamp A. et al., Am. J. Pathol. 11: 592-598 (1994). Mulligan, M.S. et al., J. Immunol. 151: 832-840 (1994) describe a similar phenomenon.

The production of humanized anti-L-selectin antibodies is described in WO 94/12215, incorporated herein by reference. The use of such antibodies in the treatment of inflammatory diseases and in particular of myocardial infarction is proposed. A dose of 1 - 50 mg is proposed to prevent acute lung failure. However, the reference does not describe a method for preventing MOF after polytrauma.

Thus, there is a need for effective treatment of preventing and/or treating multiple organ failure after polytrauma.

Acute organ damage can also be caused during cardiovascular surgery, such as an aorta-coronary vein bypass operation or a cardiac valve operation, where the blood of the patient circulates extracorporeally through a heart-lung machine. The extent of the damage is dependant on the period during which the machine is in operation. This can lead, e.g. to failure of the lungs, which can necessitate artificial respiration of the patient well after the

operation (Birnbaum, D. et al., Z. Kardiol. 79, Suppl. 4: 87-93 (1990)). Other organs such as heart, kidneys, liver or the blood and coagulation system may also be damaged and fail.

It is known from Mulligan, M.S. et al., J. Immunol. 151: 832 - 840 (1994) that molecules that promote adhesion such as for example L-, E-, and P-selectins are involved in acute inflammatory processes. These molecules mediate the adhesive interaction of leukocytes with endothelial cells. In this connection L-selectin seems to play an important role in the initial phase (rolling) of acute intrapulmonary inflammatory reactions. Mulligan states further that anti-L-selectin antibodies are suitable for shortening the duration of the lung damage that can be triggered by L-selectin.

However, up to now no preventive therapy is known which can be used to prevent acute organ damage that is caused by extracorporeal circulation of the blood. Thus, there is a need for effective treatment of preventing acute organ damage caused by extracorporeal circulation of the blood.

OBJECTS OF THE INVENTION

An object of the invention is to provide a method and a therapeutic composition which can be used to effectively prevent multiple organ failure after polytrauma in humans and to considerably reduce the mortality rate of polytrauma patients. The invention concerns the use of anti-selectin antibodies for the production of pharmaceutical compositions useful in the prevention of multiple organ failure and death after polytrauma.

An object of the invention is also to provide a method and use of a therapeutic composition which contains anti-selectin antibodies for the prevention of acute organ damage after extracorporeal circulation. Such organ damage can be largely avoided with this method and this procedure. A particular advantage of the method is the extracorporeal application which leads to an effective decrease in organ complications.

BRIEF DESCRIPTION OF THE FIGURES

Fig. 1 shows the lung wet weight of experimental animals with respect to the observation time.

Fig. 2 shows the cardiovascular parameter CO (cardiac output) with respect to time for the various experimental animals.

Fig. 3 shows the cardiovascular parameter MAP (mean arterial blood pressure) with respect to time for the experimental animals.

Fig. 4 shows the BE value (arterial base excess) with respect to time for the experimental animals.

Fig. 5 shows the number of white blood cells with respect to time for the various experimental animals.

DETAILED DESCRIPTION OF THE INVENTION

The invention concerns the use of at least one anti-selectin antibody for the production of a pharmaceutical composition to prevent acute organ damage after extracorporeal circulation of a patient's blood through a heart-lung machine, wherein 1 to 30 minutes before ending the extracorporeal circulation the anti-selectin antibody is added extracorporeally into the tube system of the heart-lung machine at a dose of 1.0 - 10 mg/kg of body weight of the patient and preferably 2 - 4 mg/kg.

Surprisingly, acute organ damage after extracorporeal circulation of a patient's blood can be prevented to a large extent by this preventive extracorporeal administration. In a preferred embodiment a total of 1 - 3 further doses of 1 - 4 mg/kg anti-selectin antibody are administered to the patient within 1 - 3 days. Polyclonal or monoclonal, murine, human, chimeric or humanized antibodies/immunoglobulins and their binding fragments can be used as the anti-selectin antibodies.

"Anti-selectin antibodies", as used herein, refers to any antibody which binds to a selectin. Especially preferred are antibodies which bind specifically to one of L-selectin, E-selectin, or P-selectin, as well as combinations of these. Also preferred ar antibides which react with more than one selectin, such as antibodies which react with both L- and E-selectin. L-selectin is a known glycoprotein that is constitutively expressed by all leukocytes. Both L-selectin and its murine homologues, GP90 and Me114, are involved in the normal recirculation of lymphocytes - each mediates the interaction between circulating lymphocytes

and vascular ligands (often referred to as "addressins") on the high endothelial venules (HEVs) of lymphoid organs (L.A. Lasky, et al., Cell 69: 927-938 (1992); E. L. Berg, et al., J. Cell Biol. 114: 343-349 (1991). In addition to its role as a lymphocyte homing receptor, L-selectin is also involved in the adhesion of circulating leukocytes to non-lymphoid tissues, such as endothelium, during inflammation. L-selectin is shed from the leukocyte surface following leukocyte activation (T.K. Kishimoto, et al., Science 245: 1238-1241 (1989)), and this may be an important process in retaining activated leukocytes at sites of inflammation. L-selectin has an amino-terminal carbohydrate-recognition domain (CRD) that has considerable homology with C-type lectins (K. Trickhamer, J. Biol. Chem. 263: 9557-7560 (1988)), followed by a single epidermal-growth-factor-like domain, complement regulatory domains, a single transmembrane polypeptide and a carboxy-terminal cytoplasmatic domain. L-selectin interacts with its cognate ligand through the amino-terminal CRD in a calcium dependent manner.

In accordance with the invention, anti-selectin antibodies are preferred which modulate, and more preferably inhibit, the interaction between the CRD domain and the corresponding carbohydrate receptors on the surface of cells. Such carbohydrate receptors are described by R.B. Parekh, Tibtech 12: 339-345 (1994), incorporated by reference. These carbohydrate receptors may be phosphorylated or sulfated sugars.

In a further embodiment of the invention, anti-P-and/or anti-E-selectin antibodies are used instead of, or in addition to, anti-L-selectin antibodies. Such antibodies can be

produced using P- or E-selectin (described in R.R. Parekh and T. F. Tedder, FASeb Journal 9: 886-873 (19995), incorporated by reference). In an especially preferred embodiment of the invention, anti-P- and/or anti-E- selectin antibodies are used which show considerable cross-reactivity with L-selectin antibodies, especially cross-reactivity with antibody HuDreg-55 or HuDreg-200.

As used herein, the term "humanized immunoglobulin" refers to an immunoglobulin comprising a human framework, at least one complementarily determining region (CDR) from a non-human antibody, and in which any constant region present is substantially identical to a human immunoglobulin constant region, *i.e.*, at least about 85-90%, preferably at least 95% identical. Hence, all parts of a humanized immunoglobulin, except possibly the CDRs, are substantially identical to corresponding parts of one or more active human immunoglobulin sequences. For example, a humanized immunoglobulin would not encompass a chimeric mouse variable region/human constant region antibody. See, e.g. European Patent Application EPA 451216, incorporated by reference.

The invention also concerns the use of such anti-selectin antibodies to reduce MOF and mortality after polytrauma. It has surprisingly turned out that it is possible to prevent multiple organ failure when anti-selectin antibodies, especially anti-L-selectin antibodies, are administered very soon after the polytrauma. This is also surprising because there are no acute symptoms at this early stage and there would therefore have been no reason to administer such a dose as a preventive measure.

It also has surprisingly turned out that anti-selectin antibodies in doses of 1.0 - 10 mg/kg, preferably of 2 - 4 mg/kg, administered one to five times, preferably once or twice after the polytraumatic event can advantageously be used, whereby the first application is given as early as possible preferably 0.5 - 8 hours and particularly preferably 0.5 - 4 hours after the polytraumatic event. The intervals between the individual applications are between about 6 and about 72 hours, preferably between 6 and 36 hours.

In a preferred embodiment the dose and time of the second and subsequent preventive applications is selected depending on the concentration of the anti-selectin antibodies in the blood and preferably in plasma or serum. In this connection it is preferable that the plasma concentration of the anti-selectin antibody is maintained at 10 - 100 μg/ml over a time period of 7 - 10 days after the polytraumatic event. This concentration is equivalent to about a 10 - 100 fold excess over the concentration of soluble selectin in plasma. The dose and time for the second and subsequent applications are determined by determining the concentration of the anti-selectin antibody in blood, serum or plasma at intervals of 6 - 24 hours and immediately administering a dose which essentially corresponds to the dose of the first application when the plasma concentration falls below 10 μg/ml antibody. When the antibody concentration is between 10 and 50μg/ml, the antibody is administered at about half the concentration of the first application, and at an antibody concentration between 50 and 100 μg/ml, no further antibody is administered. In this case only the antibody concentration is monitored further.

The anti-selectin antibody concentration in blood, serum or plasma is determined by the usual methods, preferably by an immunological method of determination. Such methods are known to a person skilled in the art. For example the determination can be carried out by means of an ELISA test in which a labelled selectin specific antibody, preferably the antibody which is also used therapeutically, competes for a specific selectin. In a subsequent step the amount of labelled antibody which has bound to the antigen is then determined and the concentration of the anti-L-selectin antibody in the sample is determined from this.

The therapeutic compositions of the invention are usually administered parenterally such as intravenously, intraarterially, intraperitoneally, subcutaneously or intramuscularly. Intravenous (i.v.) administration is preferred. The active components of the composition can be used in a liquid or solid form, preferably in a lyophilized form and be used together with a suitable diluent or carrier such as water or aqueous solutions of sodium chloride, dextrose, buffers and so forth. Other suitable pharmaceutical auxiliary substances can also be added.

Antibodies to selectin are known from the state of the art and are described for example in EP-A 0 386 906, WO 93/00111 and WO 94/12215 and by Kishimoto, T.K. et al., in Blood 78: 805 - 811 (1991) and Proc. Natl. Acad. Sci. USA 87: 2241 - 2248 (1990), all of which are incorporated by reference. L-selectin is also denoted LECAM-1, Mel 14 or Lam-1 in the literature. The cloning and sequence of Lam-1 have been described in WO 93/02698. Antibodies which bind specifically to selectin are suitable. Humanized antibodies, especially HuDreg 200 which is described in WO 94/12215 and is expressly

incorporated herein by reference, are suitable. Other antibodies which bind to L-selectin, such as HuDreg 55, (sequence: SEQ ID NO: 1 - 4) are also particularly preferred.

"Antibody" as used herein is understood as a protein that is composed of one or several polypeptide chains that are essentially encoded by antibody genes. The antibody genes code for the antigen-specific variable regions and may also code for the genes for the constant regions. Antibodies within the sense of the invention are also understood as various derivatives and fragments of antibodies such as Fv, Fab and $F(ab)_2$ and individual antibody chains (Houston et al., PNAS USA 85: 5879 - 5883 (1988), Bird et al., Science 242: 423-426 (1988), Hood et al., Immunology Benjamin N.Y., 2nd edition (1984), Hunkapiller and Hood, Nature 323: 15 - 16 (1986)). Monoclonal antibodies and fragments thereof are preferably used and particularly preferably chimeric or humanized antibodies preferably of the IgG1 or IgG4 subtype.

The antibodies preferably contain at least two light polypeptide chains and two heavy polypeptide chains. Each of these chains contains a variable region (usually the N-terminal part of the polypeptide chain) which in turn contains a domain which binds the antigen. Heavy and light chains additionally contain a constant region of the polypeptide (usually the C-terminal part) which mediates the binding of the antibody to leukocytes (neutrophils, lymphocytes etc.). Usually the light and heavy chains are complete antibody chains which are composed of the variable region and the complete constant region. In this connection, the variable regions and the constant regions can be derived from different antibodies, for

example different isotypes. For example a polypeptide which contains the variable region of a heavy chain of an anti-selectin antibody of the γ-1 isotype may be linked to the constant region of the heavy chain of an antibody from another class (or subclass).

Anti-selectin antibodies are also suitable in which one or several amino acids are substituted. In this case, amino acids are preferably substituted by other amino acids with similar characteristic features (e.g. the acidic amino acid Asp by the acidic amino acid Glu). The structural characteristics of the original sequence are not changed by such substitutions. Examples of such polypeptide structures are described in Proteins, Structures and Molecular Principles, Creighton (editor), W.H. Freeman and Company, New York (1984); Introduction to Protein Structure, C. Brandon and J. Tooze, Garland Publishing, New York (1981); Thornton et al., Nature 354: 105 (1991). In general antibodies which are suitable as anti-selectin antibodies are those which bind to one or more L-selectin, E-selectin, and P-selectin and/or inhibit the rolling of leukocytes (e.g. neutrophils).

In addition to the humanized immunoglobulins specifically described herein, other "substantially homologous" modified immunoglobulins can be readily designed and manufactured utilizing various recombinant DNA techniques well known to those skilled in the art. Human antibodies, including, for example, the Eu or GAL antibodies, as well as other human antibodies known in the art, can be used as a source of framework sequence. These framework sequences should exhibit a high degree of sequence identity with the mouse Dreg 55 or mouse Dreg 200 variable region domains from which the CDRs were derived.

The heavy and light chain variable framework regions can be derived from the same or different human antibody sequences. Indeed, the heavy and light chain framework regions can each be derived from more than one human antibody. The human antibody sequences can be the sequences of naturally occurring human antibodies or can be consensus sequences of several human antibodies. *See* Carter et al., WO 92/22653 (1992), incorporated by reference.

"Antibodies which are capable of binding in an equivalent manner" are understood as those antibodies which bind to the same or an overlapping epitope of a selectin. Epitope overlap can be determined by methods known in the art, for example with the aid of a competitive test system. A competitive binding assay may be carried out for this and the extent to which the antibody competes with, e.g., HuDreg 55 for binding to an immobilized L-selectin antigen is determined. For this, L-selectin immobilized in a suitable manner (preferably L-selectin on leukocytes) is incubated with HuDreg 55 in a labelled form and an excess of the antibody to be tested. The extent of the binding of the antibody to be tested to L-selectin is determined in comparison to HuDreg 55 by determining the bound label of the anti-leukocyte-bound label. If the labelled HuDreg 55 is displaced by at least 50 % by the antibody to be tested an epitope overlap is present. Antibodies that bind in an equivalent manner as HuDreg 55 are preferred for use in the invention.

"Antibodies which are capable of binding in an equivalent manner" can also be identified by screening for the capacity to block neutrophil-endothelial cell interaction. A

simple visual assay for detecting such interaction has been described by Kishimoto et al. (*Blood,* 78:805 (1991)). Briefly, monolayers of human umbilical vein cells are stimulated with interleukin-1. Neutrophils, with or without pretreatment with the antibody under test, are added to the monolayer under defined conditions, and the number of adhering neutrophils is determined microscopically. In one method, the neutrophils are obtained from human leukocyte adhesion deficient patients. *See* Anderson et al., *Ann. Rev. Med.* 38:175 (1987). The neutrophils from such patients lack integrin receptors, whose binding to neutrophils might obscure the effects of blocking L-selectin binding.

The antibodies can be used as complete monoclonal antibodies, fragments thereof (Fv, $(Fv)_2$, Fab', $F(ab')_2$), chimeric, humanized or human antibodies. Short antibody fragments which only contain the CDR regions or parts thereof which bind specifically to L-selectin can also be used.

The production of antibodies and in particular of monoclonal antibodies and fragments thereof is familiar to a person skilled in the art and described for example in E. Harlow and D. Lane, Antibodies: A Laboratory Manual, Cold Spring Harbor Press (1988), Bessler et al., Immunobiol. 170 (1985) 239 - 244, Jung et al., "Angewandte Chemie" 97 (1985) 883, Cianfiglia et al., Hybridoma Vol. 2 (1993) 451 - 457.

Anti-selectin antibodies that can be used according to the invention can also be produced by recombinant means. Such processes are described in Sambrook et al.,

Molecular Cloning: A Laboratory Manual, 2nd edition (1989), Cold Spring Harbor, New York, Berger and Kimmel, Methods in Enzymology, Vol. 152, Guide to Molecular Cloning Techniques (1987), Academic Press Inc., San Diego CA which are incorporated by reference. Such recombinant antibodies can be produced either in eukaryotic or prokaryotic cells by processes known to the art. Mammalian cells especially lymphocytic cell lines are preferably used as host cells. Chimeric, humanized or human antibodies are preferably produced by recombinant methodologues. Regions can be selected for the non-antigen binding regions of the antibodies which are for example described in E.A. Kabat et al., Sequences of Proteins of Immunological Interest (1987), National Institute of Health, Bethesda MD. The production of recombinant anti-L-selectin antibodies and in particular of humanized and human antibodies is described in WO 94/12215, which is hereby incorporated by reference. A particularly preferred, humanized anti-L-selectin antibody is HuDreg 55, which may be constructed in the same manner as HuDreg 200 described therein, and comprises two light chains having the sequence SEQ ID NO: 2 and two heavy chains having the sequence SEQ ID NO: 4.

Preferred humanized immunoglobulins are those which bind to selectin with a binding affinity of at least 1 x 10^7 M^{-1} in standard binding conditions (*e.g.*, phosphate-buffered saline with 2 percent fetal bovine serum at 25°C). Examples of such humanized immunoglobulins are Dreg 55 and Dreg 200. More preferred are humanized antibodies, which preferably bind, in standard binding conditions, to human selectin with an affinity of at least 1 x 10^8

M^{-1}, and more preferably, with an affinity of at least 1 x 10^9 M^{-1}, and advantageously with an affinity of at least 1 x 10^{10} M^{-1} or more. Usually, the binding affinity of a humanized immunoglobulin is within a factor of 3-10 of the mouse immunoglobulin from which it was derived. For example, the affinity of the mouse Dreg 200 antibody is about 10^8 M^{-1} and that of mouse Dreg 55 is about

10^9 M^{-1}.

The following examples, sequence protocols, publications and figures further elucidate the invention. The described processes are to be understood as examples of illustration, not of limitation, which also after modifications, describe the subject matter of the invention.

EXAMPLE 1 - Use Of Anti-L-Selectin Antibody To Reduce Post-Trauma Organ Failure

The protective action of a humanized antibody against L-selectin (anti-L-selectin) in reducing post-traumatic organ failure such as that which typically occurs after injury in patients with severe polytrauma is demonstrated. Humanized anti-L-selectin antibody (HuDreg 55) is used as the antibody. It also reacts with baboon L-selectin. This mouse form of this antibody is described by Kishimoto PNAS, USA 87 (1990) 2244 - 2248. The humanized sequence is shown in SEQ ID NO: 1 - 4.

The HuDreg 55 and HuDreg 200 antibodies react with L-selectin on human leukocytes; however, only HuDreg 55 reacts with L-selectin of baboon leukocytes. Therefore HuDreg 55 was used. Since HuDreg 55 and HuDreg 200 bind in the same

concentration range to human leukocytes (e.g. in FACS analysis), the effects of HuDreg 55 on baboons is presumptively equivalent to the effect of HuDreg 200.

As a model, severe tissue damage with associated hypovolemia (= loss of liquid and blood towards the inside and/or outside) was induced in baboons. The pure blood loss with subsequent shock (hemorrhagic shock) is less relevant for the lung damage (Pretorius et al., J. Trauma 1987; 27: 1344 - 1353; Schlag et al., page 384-402, in Schlag, Redl: Pathophysiology of Shock, Sepsis, and Organ Failure, Springer Verlag, Berlin, 1993). This is in agreement with clinical experience which shows that lung complications only occur very rarely in pure hemorrhagic shock (Schlag et al., 1993 see above).

In order to determine the frequency and severity of post-traumatic lung failure it was necessary to observe the animals (named: SELEC 971, SELEC 979 (treated); and Co 968, Co 969, Co 970 (control)) over several days; however, for ethical reasons, it was not possible to induce bone fractures in conscious animals and leave them untreated for several days so that in this subchronic model the tissue trauma is simulated. The activation of the complement system appears to be the earliest trigger for the activation of cellular systems and plays a key role in the rapid occurrence of a non-bacterial inflammatory reaction of the body (Schlag et al., 1993, supra). Therefore, in the model, complement was activated by cobra venom factor. The mortality for multiple organ failure after severe polytrauma was quoted as 15 - 30 %. In the present animal model the severity of the polytrauma was

increased to the extent that the mortality is at least twice as high and occurred earlier than in humans. Therefore the observation period was limited to three days.

Adult baboons with a body weight (BW) between 18 and 22 kg were admitted to the study after three months quarantine. The fasted animals were sedated with ketamine (6-8 mg/kg), subsequently intubated and attached to a CPAP respirator (continuous positive airway pressure) (inspiratory O_2 concentration of 25 $\pm$ 2%). The anesthesia was maintained with 1-3 mg/kg/h pentobarbital. The animals breath spontaneously. A Swan Ganz catheter was pushed forward into the pulmonary artery via the right femoral vein. A catheter for withdrawing blood and measuring blood pressure was tied into the right arm artery. A large lumen catheter is introduced into the femoral artery for the temporary collection of blood. A catheter for infusions, medication and blood collection was introduced into the left arm vein. The bladder was catheterized for the measurement of urine production. The Swan Ganz catheter and the arterial catheter were left for three days. For fluid balance the animals received 5 ml/kg/h Ringer solution (electrolyte solution for parenteral liquid substitution) during the anaesthetic phase. The blood temperature of the animals was kept at $\geq$ 37°C with the aid of an infrared lamp. Blood gas analyses were carried out (pO_2, pCO_2, pH, BE, HCO^-_3) and hemodynamic parameters were determined (MAP, RAP, PAP, CO, HR). Lung function was determined by means of the respiratory rate (RR) and end expiratory CO_2. Blood samples were collected repeatedly in order to measure the number of white blood cells (WBCs). Cobra venom factor was administered at a dose of 10 U/kg per i.v. at the beginning of the retransfusion and administered again at a dose of 5 U/kg 1 hour after

beginning the retransfusion of the blood. The blood withdrawal for triggering the hypovolemia was regulated such that the MAP (mean arterial pressure) comes to lie between 40 and 50 mm Hg and the CO (cardiac output) is reduced by 50 to 70%. Approximately 50 ml/kg blood were usually withdrawn for this and stored until retransfusion. The deficient circulation was maintained for two to three hours and was controlled in such a way that the base excess was no more than -5 to -7 mEq. At the end of this shock phase retransfusion of the previously collected blood was begun. This phase lasted 4 hours.

The retransfusion was complemented by an additional administration of Ringer solution. Humanized antibody HuDreg 55 or the corresponding volume of saline solution as a placebo was administered intravenously 15 minutes after the start of retransfusion. Anti-L-selectin antibody was administered at a dose of 2 mg/kg. At the end of retransfusion the animals were awakened from anaesthesia and they were returned to their cages for observation. At times 24 h, 48 h and 72 h a low level of anaesthesia was again induced and the measuring parameters were registered and blood was withdrawn. If the animals had not died before the end of the three day observation period, they were then sacrificed and autopsied. The main terminal points of the study were mortality, survival period and organ damage, for example to the lung.

In the first experiment, three control animals were treated with placebo solution and two with the HuDreg 55 humanized antibody. Of the three control animals, two died before the end of the three day observation period at 38 h and 41 h whereas both anti-L-selectin

treated animals survived. The lung wet weight, as an expression of organ damage, was almost normal in the antibody animals (normal values 7-8 g/kg BW) whereas it had increased considerably in all three placebo-treated control animals (Fig. 1). This is due to infiltration of fluid after the permeability disorder. The cardiovascular parameters CO and MAP (Fig. 2 and 3) are better at 24 hours in the surviving animals than in the control animals. The dying control animals also have a negative arterial base excess (BE) indicating a disturbed acid-base balance (Fig. 4). The leucocytosis (increase in white blood cells) observed in the control animals is absent in the antibody animals (Fig. 5).

EXAMPLE 2 - Use Of Anti-L-Selectin Antibody To Reduce Post-Traumatic Mortality

The experiments reported in Example 1, supra, were continued and expanded to include 28 baboons which were randomly assigned to one of two experimental groups conducted as described in Example 1. The baboons received either 2 mg/kg i.v. of anti-L-Selectin antibody or the appropriate placebo volume-dose as control 15 minutes after initiation of reperfusion after the ischemia period. The main endpoints for statistical analysis of the study were mortality at the end of the 3-day observation period and survival time. Fisher's exact test was used for mortality analysis and the log-rank-test was used for survival time analysis. One-sided p-values (reduction of mortality or prolongation of survival time by active treatment) are reported. The null hypothesis was rejected only when the probability (p) of the calculated statistic was $p<0.05$.

Anti-L-Selectin antibody reduced ($p<0.05$) mortality from 10 out of 14 (=71%) baboons in the control group to 3 out of 14 (=21%) baboons in the active treatment group at a level of statistical significance. In addition, survival time in the anti-L-Selectin group was prolonged to 64.4 h, whereas animals in the control group died earlier ($p<0.05$), on an average at 42.1 h. This difference was statistically significant.

The table summarizes the results:

	Mortality	Survival time (h)
Anti-L-Selectin Antibody	3/14*	$64.4 \pm 4.7^{+}$
Placebo-control	10/14	42.4 ± 5.7

Mean $\pm$ standard error of mean;
n = 14 per group;
*, $p < 0.05$ by Fisher's exact test;
$^{+}$, $p<0.05$ by log-rank-test.
Observation period was 72 h.

These data show that early treatment of baboons suffering from ischemia-reperfusion injuries due to hemorrhagic-traumatic shock with administration of anti-L-Selectin significantly prolongs survival time and reduces mortality as compared to placebo-control.

EXAMPLE 3 - Use of Anti-L-Selectin Antibody To Reduce Organ Damage After Extracorporeal Blood Circulation

The protective action of a humanized antibody against L-selectin, preferably HuDreg 55, in reducing organ damage after extracorporeal blood circulation such as that which

typically occurs after long operating periods of the heart-lung machine in cardiac surgery was studied.

As a model, severe lung damage was caused in baboons by letting the heart-lung machine, which takes over the function of the lungs and heart after the heart is stopped, run for several hours. After the machine was turned off the pumping action of the heart was resumed and endogenous circulation and respiration restarted, massive infiltration of activated leukocytes into the pulmonary circulation caused severe damage of the lungs. The leukocytes present in the pulmonary circulation locally release toxic mediators at a high concentration which leads to damage of the vascular endothelium with subsequent increase in permeability. In this process fluid crossed over from the vascular space into the alveoli (smallest pulmonary alveoli) which led to an accumulation of fluid in the lung. This impeded gas exchange in the lung and artificial respiration becomes necessary. The oxygen demand increases as the impairment in gas exchange increases in severity and this was further aggravated by a fibroproliferative transformation of the alveolo-endothelial barrier. Thus, in particularly severe cases, the concentration of inhaled oxygen in the respiratory air which is usually about 20 % has to be increased to about 100 %. Nevertheless, in such cases, the supply of pure oxygen is insufficient to maintain the arterial oxygen concentration or oxygen partial pressure in the blood (paO_2) at an adequate level.

The fibroproliferative transformation process and pulmonary edema result in an increase in the pressures in the arteria pulmonalis which is connected to the lung and this

leads to a strain on the right heart. If these reactions build up further this finally leads to death by heart-lung failure.

Adult baboons with a body weight (BW) between 18 and 22 kg are admitted to the study after three months quarantine. The fasted animals were sedated with ketamine (6-8 mg/kg), subsequently intubated and attached to a CPAP respirator (inspiratory O_2 concentration of 25 $\pm$ 2%). The anesthesia was maintained with 1-3 mg/kg/h pentobarbital. The animals breathed spontaneously. A Swan Ganz catheter was pushed forward into the pulmonary artery via the right femoral vein. A catheter for withdrawing blood and measuring blood pressure was tied into the right arm artery. A catheter for infusions, medication and blood collection was introduced into the left arm vein. The bladder is catheterized to measure the production of urine. For fluid balance the animals receive 5 ml/kg/h Ringer solution. The temperature of the animals is maintained at 37°C with the aid of an infrared lamp. Blood gas analyses are carried out (pO_2, pCO_2, pH, BE, HCO^-_3) and hemodynamic parameters are determined (MAP, RAP, PAP, CO, HR). The lung function is determined by means of the respiratory rate (RR) and end expiratory CO_2. Blood samples are collected repeatedly in order to measure the number of white blood cells (WBC).

At the start of the experiment the thorax was opened (thoracotomy) and the vena cava and the aorta was prepared. Afterwards, firstly the vena cava and then the aorta were cannulated so that blood from the vena cava flows into the heart-lung machine and later back into the aorta. A peristaltic pump assumes the pumping function of the heart in the

heart-lung machine and ensures maintenance of the pressure gradient required for the circulation. Exchange of oxygen and binding of carbon dioxide is achieved by membrane oxygenation. The blood was heparinized so that the tubes and blood vessels do not get blocked. The blood flows back to the aorta via the tube system and is distributed in the body via the normal vascular system.

The heart-lung machine takes over the function of the heart and lung. The heart is stopped while the machine is in operation so that the operating surgeon can for example work on the cardiac valves (insert prostheses).

Fifteen minutes before the end of the four hour extra-corporeal circulation, a dose of 2 mg/kg HuDreg 55 or the same volume dose of placebo was administered directly into the tube system of the heart-lung machine. The animal was observed for a further four hours after ending the extracorporeal circulation. Measurements are carried out repeatedly before, during and after the extra-corporeal circulation. In particular arterial blood gases and parameters for acid-base balance are recorded, cardiovascular parameters such as the mean arterial blood pressure, right atrial pressure, pulmonary artery pressure, cardiac output and heart rate are determined, the lung function is measured (e.g. end expiratory CO_2) and blood samples are withdrawn for hematological, clinical-chemical (e.g. kidney and liver function) and biochemical analyses. In addition urine production (kidney function) is measured. Further parameters for permeability disorders in the lung were determined. At the end of the experiment the animals were sacrificed and necropsy and histological examinations were

carried out in order to determine the degree of damage to the various organs such as heart, lung, liver, kidney, intestine, CNS, blood etc. It is expected that the animals treated with HuDreg 55 sustain less organ damage than those treated with placebo.

SEQUENCE LISTING

(1) GENERAL INFORMATION:

(i) APPLICANTS:
(A) NAME: Martin, Ulrich, et al.

(ii) TITLE OF INVENTION: Anti-L-selectin antibodies for prevention of multiple organ failure after polytrauma and for prevention of acute organ damage after extracorporeal blood circulation.

(iii) NUMBER OF SEQUENCES: 4

(iv) CORRESPONDENCE ADDRESS:
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(C) New York
(D) New York
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(F) 10022

(v) COMPUTER READABLE FORM:
(A) MEDIUM TYPE: 3.5" Computer Disk
(B) COMPUTER: IBM PC compatible
(C) OPERATING SYSTEM: PC-DOS/MS-DOS
(D) SOFTWARE: ASCII

(vi) CURRENT APPLICATION DATA:
(A) APPLICATION NUMBER: To be assigned
(B) 20-Dec-95
(C) CLASSIFICATION: 530

(vii) PRIOR APPLICATION DATA:
(A) APPLICATION NUMBER: EP 95 112 895.8
(B) FILING DATE: 17-Aug-95

(vii) PRIOR APPLICATION DATA:
(A) APPLICATION NUMBER: EP 95 114 969.9
(B) FILING DATE: 19-Sep-95

(vii)PRIOR APPLICATION DATA:
(A) APPLICATION NUMBER: US 08 578 953
(B) FILING DATE: 27-Dec-1995

(viii) ATTORNEY/AGENT INFORMATION
(A) NAME: Hanson, Norman D.
(B) REGISTRATION NUMBER: 30.946
(C) REFERENCE/DOCKET NUMBER: BOER 1059-PFF/NDH

(ix) TELECOMMUNICATION INFORMATION
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(B) TELEFAX: (212) 838-3884

(2) INFORMATION FOR SEQ ID NO: 1:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 654 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: double
(D) TOPOLOGY: linear

(ii) MOLECULE TYPE: cDNA

(ix) FEATURE:
(A) NAME/KEY: CDS
(B) LOCATION: 1,,654

(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 1:

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165 170 175

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180 185 190

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His Lys Val Tyr Ala Cys Glu Val Thr His Gln Gly Leu Ser Ser Pro
195 200 205

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210 215

(2) INFORMATION FOR SEQ ID NO: 2:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 218
(B) TYPE: amino acid
(C) STRANDEDNESS: single
(D) TOPOLOGY: linear

(ii) MOLECULE TYPE: protein

(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 2:

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50 55 60

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165 170 175

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195 200 205

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(2) INFORMATION FOR SEQ ID NO: 3:
(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 1329 base pairs
(B) TYPE: nucleic acid
(C) STRANDEDNESS: double
(D) TOPOLOGY: linear
(ii) MOLECULE TYPE: cDNA

(ix) FEATURE:
(A) NAME/KEY: CDS
(B) LOCATION:1,,1329
(ix) FEATURE:
(A) NAME/KEY: mat_peptide
(B) LOCATION:1

(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 3:

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100 105 110

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CCC TGC TCC AGG AGC ACC TCC GAG AGC ACA GCC GCC CTG GGC TGC CTG 432
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145 150 155 160

GCC CTG ACC AGC GGC GTG CAC ACC TTC CCG GCT GTC CTA CAG TCC TCA 528
Ala Leu Thr Ser Gly Val His Thr Phe Pro Ala Val Leu Gln Ser Ser
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GGA CTC TAC TCC CTC AGC AGC GTG GTG ACC GTG CCC TCC AGC AGC TTG 576
Gly Leu Tyr Ser Leu Ser Ser Val Val Thr Val Pro Ser Ser Ser Leu
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Gly Thr Lys Thr Tyr Thr Cys Asn Val Asp His Lys Pro Ser Asn Thr
195 200 205

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225 230 235 240

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435 440

(2) INFORMATION FOR SEQ ID NO: 4:

(i) SEQUENCE CHARACTERISTICS:
(A) LENGTH: 443
(B) TYPE: amino acid
(C) STRANDEDNESS: double
(D) TOPOLOGY: linear

(ii) MOLECULE TYPE: protein

(xi) SEQUENCE DESCRIPTION: SEQ ID NO: 4:

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35 40 45

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50 55 60

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Asn Val Phe Ser Cys Ser Val Met His Glu Ala Leu His Asn His Tyr
420 425 430

Thr Gln Lys Ser Leu Ser Leu Ser Leu Gly Lys
435 440

WHAT IS CLAIMED IS:

1. A method for preventing acute organ damage associated with extracorporeal circulation of a patient's blood through a heart-lung machine, comprising contacting the patient's blood circulating through the heart-lung machine with a pharmaceutical composition comprising at least one anti-selectin antibody in a pharmaceutically acceptable carrier, said pharmaceutical composition being contacted to the patient's blood 1 - 30 minutes before ending the extracorporeal circulation and at a dose of 1.0 - 10 mg/kg of body weight of the patient.

2. The method of claim 1, wherein said anti-selectin antibody is an anti-L-selectin antibody.

3. The method of claim 1, wherein said anti-selectin antibody is an anti-E-selectin antibody.

4. The method of claim 1, wherein said anti-selectin antibody is an anti-P-selectin antibody.

5. The method according to claim 1, wherein said pharmaceutical composition is administered at a dose of 2 - 4 mg/kg of body weight of the patient.

6. The method according to claim 1, wherein the anti-selectin antibody is a humanized monoclonal antibody.

7. The method according to claim 1, wherein the anti-L-selectin antibody is HuDreg 200.

8. The method according to claim 1, wherein the anti-L-selectin antibody is HuDreg 55.

9. The method according to claim 1, further comprising administering directly to the patient an additional 1 - 3 doses of the pharmaceutical composition at 1 - 4 mg/kg of body weight of the patient for 1-3 days.

10. A method for treating a patient who has suffered a polytraumatic event, comprising administering a therapeutically effective amount of a dose of an anti-selectin antibody in a pharmaceutically acceptable carrier to said patient.

11. The method of claim 10, wherein said anti-selectin antibody is an anti-L-selectin antibody.

12. The method of claim 10, wherein said anti-selectin antibody is an anti-E-selectin antibody.

13. The method of claim 10, wherein said anti-selectin antibody is an anti-P-selectin antibody.

14. The method of claim 10, wherein a dose ranging from 1.0 - 10 mg/kg of body weight of the patient of the anti-selectin antibody in a pharmaceutically acceptable carrier is administered 1 - 5 times after the polytraumatic event.

15. The method of claim 10, wherein the first dose is administered 0.5 - 8 hours after the polytraumatic event.

16. The method of claim 15, wherein the first dose is administered 0.5 - 4 hours after the polytraumatic event.

17. The method of claim 14, wherein the interval between administration of the doses of the anti-selectin antibody in a pharmaceutically acceptable carrier ranges between 6 and 72 hours.

18. The method of claim 17, wherein the interval between administration of the doses of the anti-selectin antibody in a pharmaceutically acceptable carrier ranges between 6 and 36 hours.

19. The method of claim 10, wherein doses of the anti-selectin antibody and a pharmaceutically acceptable carrier are administered up to 10 days after the polytraumatic event, and the concentration and time of administration of the doses is determined by the concentration of the anti-selectin antibody in the serum or plasma of the patient at intervals of 6 - 24 hours after administration of the previous dose, wherein when

(a) the concentration of said anti-selectin antibody is less than 10 μg/ml of said patient's serum or plasma, then a dose at least as high the previous dose, up to a maximum dose 10 mg/kg, is administered, or when

(b) the concentration of said anti-selectin antibody is between 10 μg/ml and 50 μg/ml of said patient's serum or plasma, then a dose which is half that of the previous dose is administered, or when

(c) the concentration of said anti-selectin antibody is greater than 50 μg/ml of said patient's serum or plasma, then a dose of anti-L-selectin antibody and a pharmaceutically acceptable is not administered, and the patient's serum or plasma is further monitored in accordance with steps (a) and (b).

20. A method of claim 10, wherein the anti-selectin antibody is a humanized antibody.

21. The method of claim 20, wherein the anti-selectin antibody is HuDreg 55 or HuDreg 200.

22. A method for reducing the probability of incidence of organ failure after a polytraumatic event, comprising administering an amount of an anti-selectin antibody in a pharmaceutically acceptable carrier to said patient, in an amount sufficient to reduce probability of incidence of said organ failure.

23. The method of claim 22, wherein the anti-selectin antibody is humanized.

24. The method of claim 22, wherein said anti-selectin antibody is an anti-L-selectin antibody.

25. The method of claim 22, wherein said anti-selectin antibody is an anti-P-selectin antibody.

26. The method of claim 22, wherein said anti-selectin antibody is an anti-E-selectin antibody.

27. The method of claim 22, wherein the anti-L-selectin antibody is Dreg 55 or HuDreg 55.

28. The method of claim 22, wherein said organ failure is multiple organ failure.

ABSTRACT

Anti-selectin antibodies for reducing probability of incidence of polytraumatic events, such as organ failure.

Fig. 1
Lung wet weight (at the time of death stated below)
Polytrauma study on the baboon

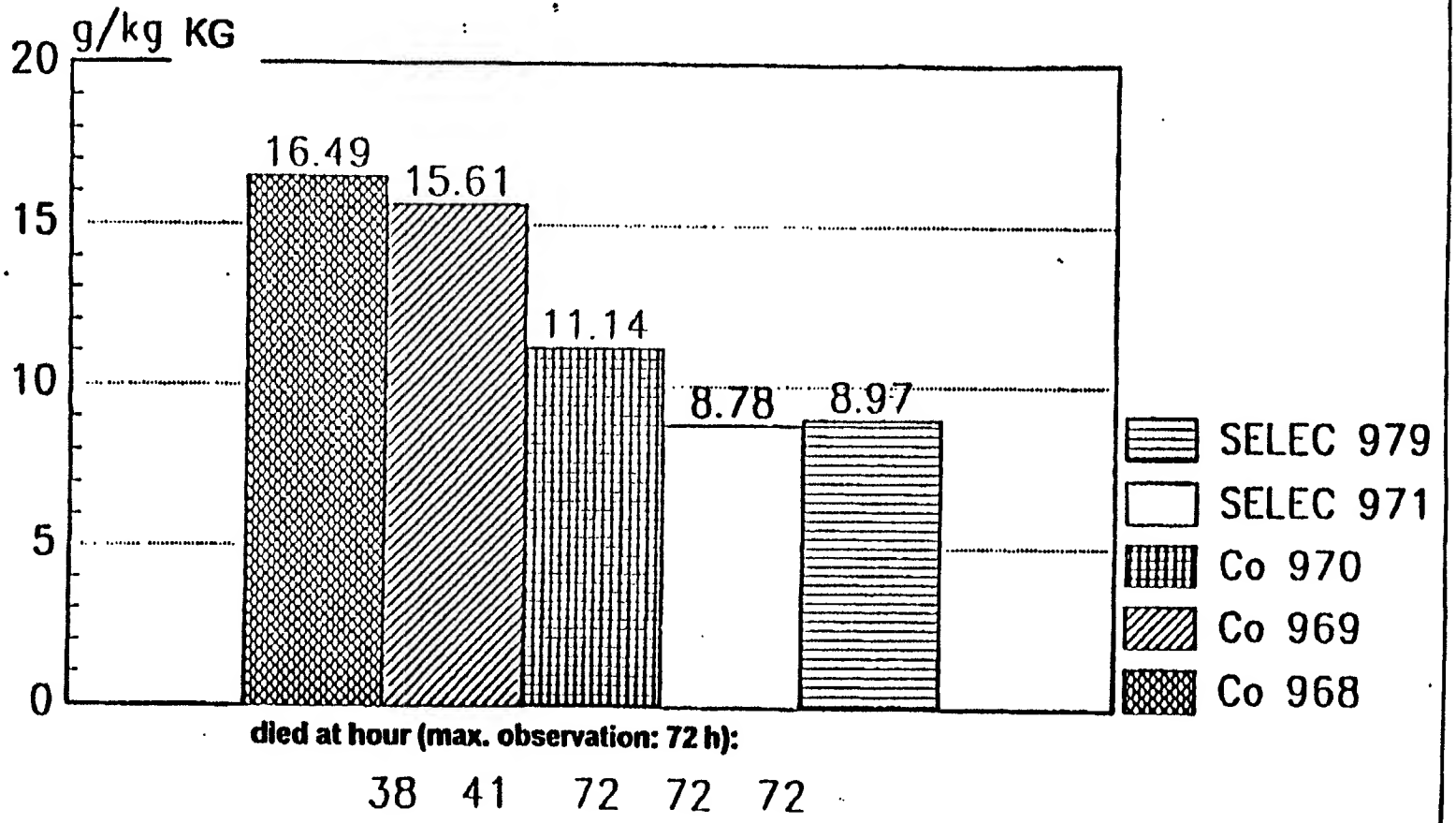


Fig. 2

Cardiac output

Polytrauma study on the baboon

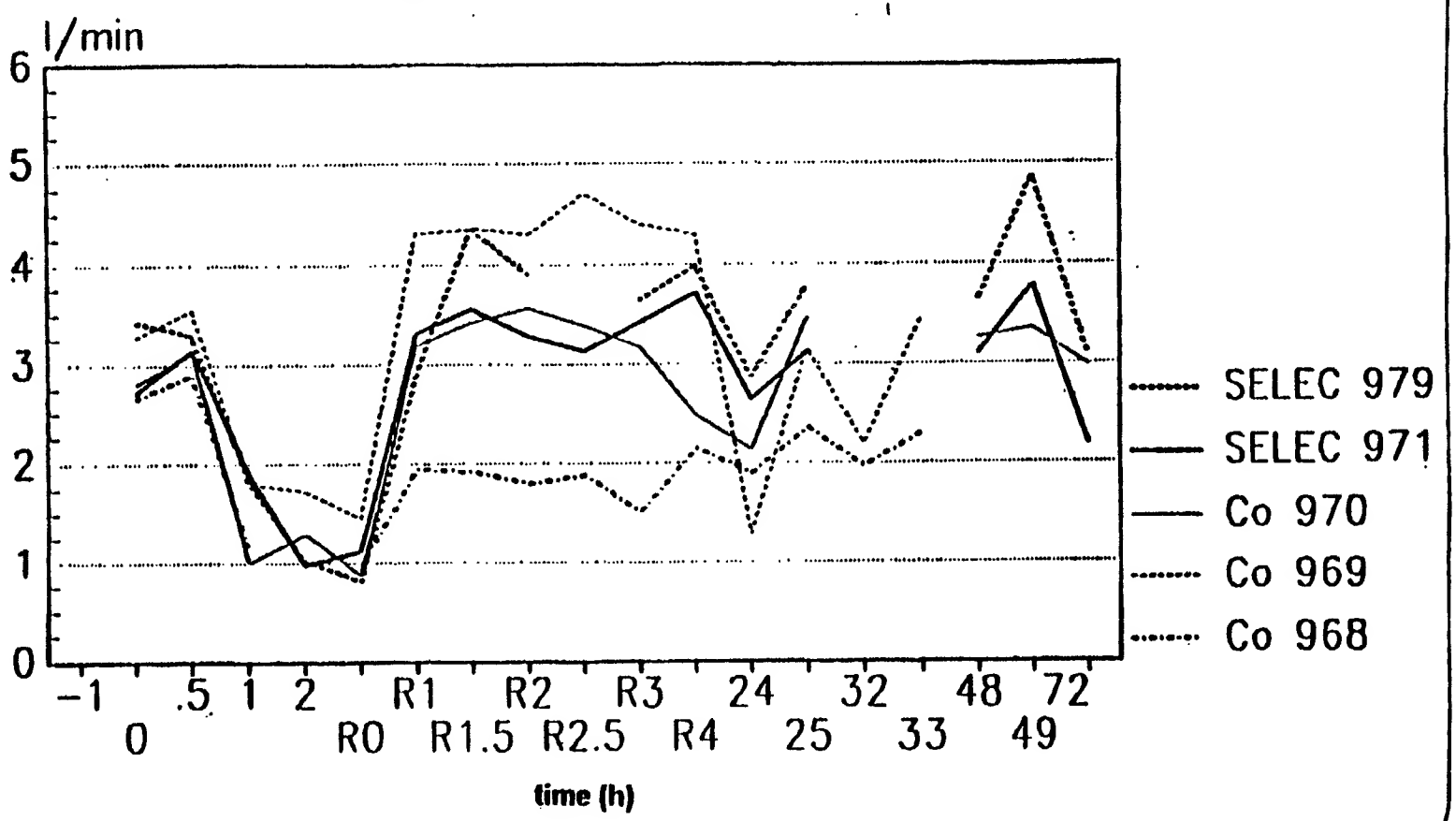


Fig. 3

Mean arterial blood pressure

Polytrauma study on the baboon

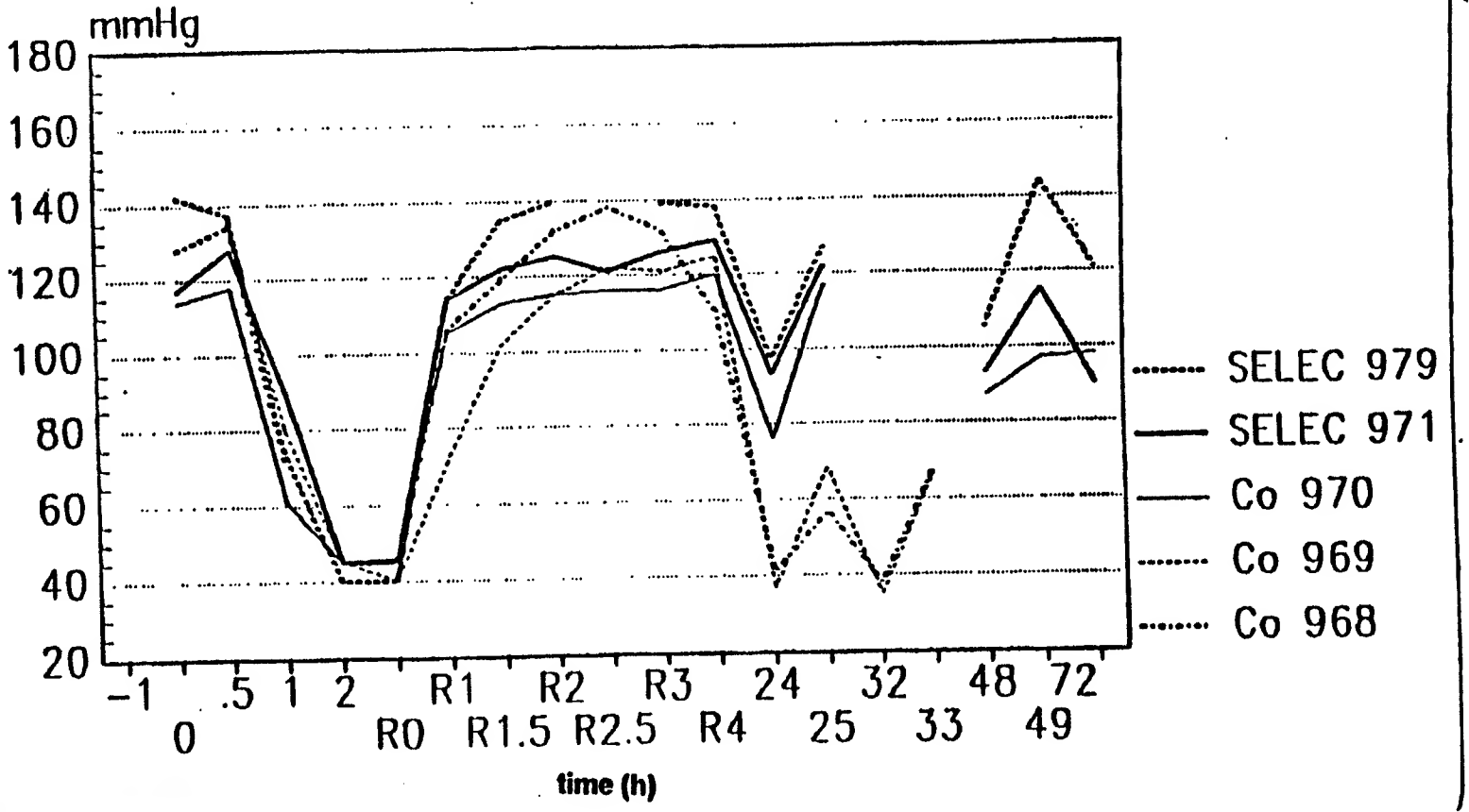
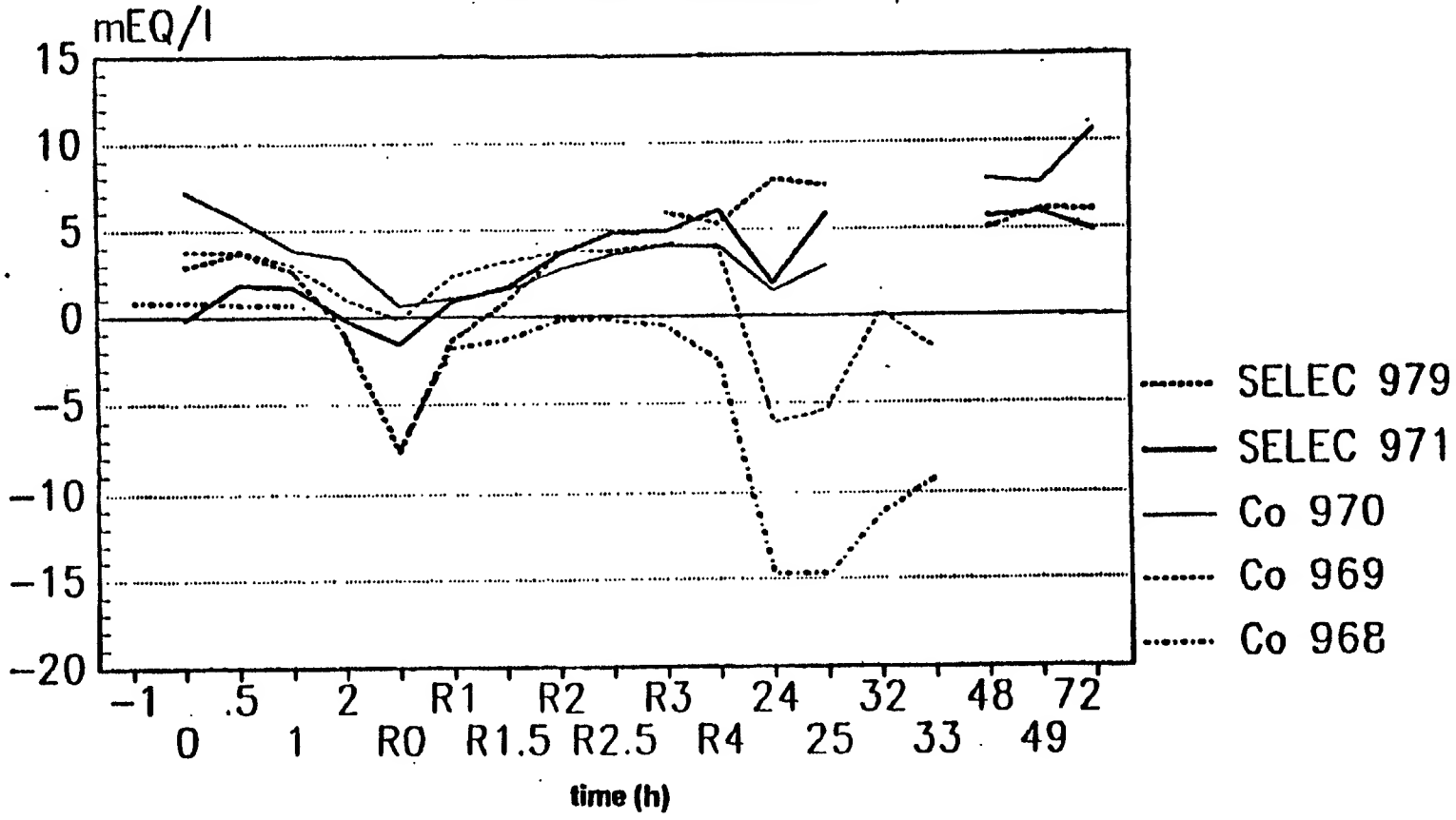


Fig. 4
Arterial base excess
Polytrauma study on the baboon



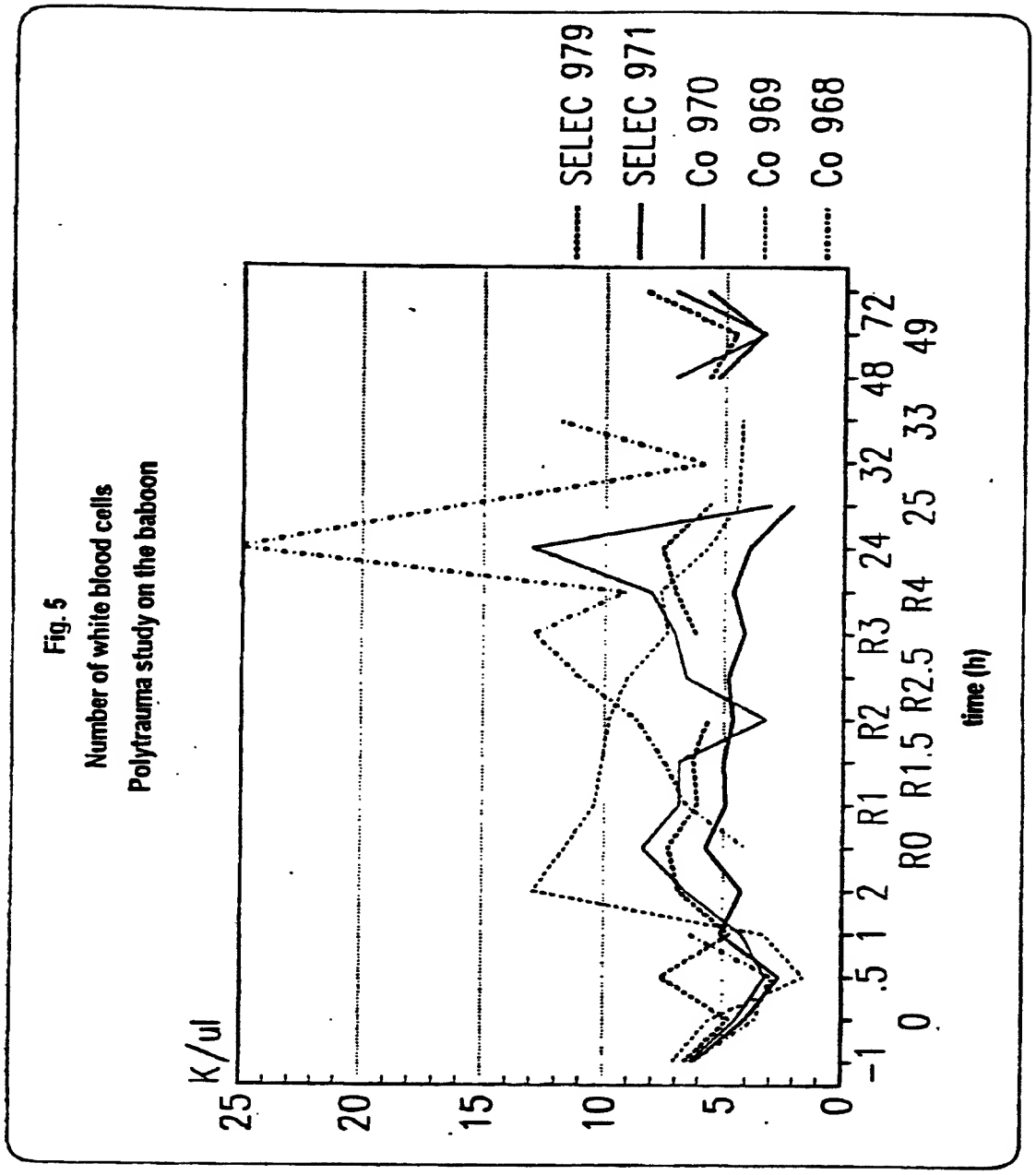


Fig. 5
Number of white blood cells
Polytrauma study on the baboon
K/ul
25
20
15
10
5
0
-1
.5
1
2
R0
R1
R1.5
R2
R2.5
R3
R4
24
25
32
33
48
49
72
time (h)
SELEC 979
SELEC 971
Co 970
Co 969
Co 968

DECLARATION/POWER OF ATTORNEY

As a below named inventor, I hereby declare that:

My resident, post office address and citizenship are as stated below next to my name.

I believe I am the original, first and sole inventor (if only one name is listed below) or an original, first and joint inventor (if plural names are listed below) of the subject matter which is claimed and for which a patent is sought on the invention entitled ANTI-SELECTIN ANTIBODIES FOR PREVENTION OF MULTIPLE ORGAN FAILURE AFTER POLYTRAUMA AND FOR PREVENTION OF ACUTE ORGAN DAMAGE AFTER EXTRACORPOREAL BLOOD CIRCULATION, the specification of which

(X) is attached hereto.
( ) was filed on _____ as Application Serial No. _____
and was amended on (1) _____, (2) _____ (if applicable).

I hereby state that I have reviewed and understand the contents of the above identified specification, including the claims, as amended by any amendment referred to above.

I acknowledge the duty to disclose information which is material to the examination of this application in accordance with Title 37, Code of Federal Regulations, 1.56(a).

Foreign Priority Applications

I hereby claim foreign priority benefits under Title 35, United States Code 119 of any foreign application(s) for patent or inventor's certificate listed below and have also identified below any foreign application for patent or inventor's certificate having a filing date before that of the application on which priority is claimed:

			Priority Claimed
PCT/US96/13152 (Number)	*PCT Application* (Country)	*13 August 1996* (Day/Month/Year Filed)	*Yes (X) No ( )*
_____ (Number)	_____ (Country)	_____ (Day/Month/Year Filed)	*Yes ( ) No ( )*

U.S. Priority Applications

I hereby claim the benefit under Title 35, United States Code, §120 of any United States application(s) listed below and, insofar as the subject matter of each of the claims of this application is not disclosed in the prior United States application in the manner provided by the first paragraph of Title 35, United States Code, §112, I acknowledge the duty to disclose material information as defined in Title 37, Code of Federal Regulations, §1.56(a) which occurred between the filing date of the prior application and the national or PCT international filing date of this application:

08/578,953 (Applic. Serial No.)	*December 27, 1995* (Filing Date)	*Pending* (Status-patented/pending/abandoned)
_____ (Applic. Serial No.)	_____ (Filing Date)	_____ (Status-patented/pending/abandoned)

Power of Attorney

I hereby appoint the following attorneys to prosecute this application and transact all business in the Patent and Trademark Office connected therewith: John E. Lynch, Reg. No. 20,940; Peter F. Felfe, Reg. No. 20,297; Norman D. Hanson, Reg. No. 30,946; Andrew L. Tiajoloff, Reg. No. 31,575; John A. Bauer, Reg. No. 32,554; Anne Schofield, Reg. No. 36,669; Madeline F. Baer, Reg. No. 36,437 and James R. Crawford, Reg. No. 39,155, my attorneys with full power of substitution and revocation. Address all telephone calls to Norman D. Hanson, at (212) 688-9200. Address all correspondence to:

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I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

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